SUMMARY BASIS OF APPROVAL

Reference No:
90-0566

Proper Name:
Antibody to Hepatitis B Surface Antigen

Applicant:
Abbott Laboratories
1 Abbott Park Rd.
Abbott Park, IL 60064

Product Trade Names:
IMx® HBsAg
IMx® HBsAg Confirmatory

I. INDICATIONS FOR USE

IMx HBsAg is an in vitro qualitative third generation Microparticle Enzyme Immunoassay (MEIA) for the detection of Hepatitis B Surface Antigen (HBsAg) in human serum or plasma.

The primary purpose of this assay is to screen blood donations so that units intended for transfusion containing HBsAg can be identified and eliminated from the blood supply.

II. BRIEF DESCRIPTION OF TEST

The IMx HBsAg assay utilizes a latex microparticle solid phase coated with mouse monoclonal anti-HBs antibodies to capture HBsAg from human sera or plasma. Captured HBsAg is labeled with a biotin tag by reaction with goat anti-HBs/biotin conjugate. These three components, sample, microparticles and biotin conjugate, are incubated simultaneously in the first incubation step to form a biotin labeled complex. The biotin labeled complex is detected by incubation with rabbit anti-biotin coupled to alkaline phosphatase in a second step. Bound enzyme is detected fluorometrically. The intended use of this assay is the screening of human sera and plasma for the presence of HBsAg.

Confirmation of IMx HBsAg reactive specimens is accomplished by specific antibody neutralization with Human antibody to Hepatitis B Surface Antigen (anti-HBs). The IMx Confirmatory Reagents consist of a neutralizing reagent (Reagent A) and a nonreactive, nonneutralizing control reagent (Reagent B). The assay consists of an off-line neutralization where each sample (both neat and at a 1:500 dilution), the Mode 1 Calibrator and Positive Control are pipetted into the sample well of two reaction cells. Reagent A is added to one reaction cell and Reagent B is added to the other. After the preincubation period, the sample mixtures are tested per the IMx HBsAg assay. Neutralization is determined by a comparison of the neutralization reagent result with the control reagent result. Specimens reactive in the IMx HBsAg assay that meet the criteria for neutralization are considered confirmed positive for the presence of HBsAg.

IMx HBsAg consists of the following components:

Contained in the IMx HBsAg Reagent Pack (1. thru 4. below only):

1. 1 bottle (3.0 mL) Anti-HBs (Mouse, Monoclonal, IgM) Coated Microparticles in buffer containing Porcine Skin Gelatin, Minimum concentration: 0.15% (w/v) microparticles. Preservative: 0.2% Sodium Azide.

2. 1 bottle (8.5 mL) Anti-Biotin (Rabbit): Alkaline Phosphatase Conjugate in TRIS Buffer with (Rabbit) IgG. Minimum concentration: 0.03 µg/mL. Preservative: 0.1% Sodium Azide.

3. 1 bottle (10 mL) 4-Methylumbelliferyl Phosphate, 1.2 mM, in buffer. Preservative: 0.1% Sodium Azide.

4. 1 bottle (13.5 mL) Biotinylated Anti-HBs (Goat, IgG) in buffer containing animal sera (Goat, Calf, Rabbit, Mouse). Minimum concentration: 0.25 µg/mL. Preservative: 0.1% Sodium Azide, 0.1% Nipasept®, and 0.0005% Quinolone.

- 1 bottle (6 mL) IMx HBsAg MODE 1 Calibrator. Recalcified human plasma nonreactive for HBsAg, anti-HBs, anti-HCV and anti-HIV-1/HIV-2. Concentration: 0 ng/mL. Preservative: 0.1% Sodium Azide. Dye: Green (Acid Yellow No. 23 and Acid Blue No. 9).

- 1 bottle (8 mL) IMx HBsAg Positive Control. Inactivated human plasma reactive for HBsAg diluted in recalcified human plasma nonreactive for HBsAg, anti-HBs, anti-HCV and anti-HIV-1/HIV-2. Preservative: 0.1% Sodium Azide. Dye: Bromophenol Blue.

- 1 bottle (8mL) IMx HBsAg Negative Control. Recalcified human plasma nonreactive for HBsAg, anti-HBs, anti-HCV and anti-HIV-1/HIV-2. Preservative: 0.1% Sodium Azide.

The IMx HBsAg Controls have the following ranges:

Control	Color	Concentration	Range (S/N)
Negative	Natural	0 ng/mL	0.75 - 1.30
Positive	Blue+	4 - 15 ng/mL	7.00 - 63.00

+Dye: Bromophenol Blue

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Other Reagents:

- 1 bottle (110 mL) IMx Probe Cleaning Solution containing 2% Tetraethylammoniumhydroxide (TEAH) in distilled water.

- 4 bottles (1000 mL each) IMx MEIA #2 Diluent Buffer containing 0.3M Sodium Chloride in TRIS Buffer. Preservative: 0.1% Sodium Azide and Antimicrobial Agents.

IMx HBsAg Confirmatory consists of the following components:

- 1 bottle (1 mL) REAGENT A. Antibody to Hepatitis B Surface Antigen (Human) nonreactive for HBsAg and anti-HIV-1/HIV-2. Minimum concentration (anti-HBs): 430 mIU/mL. Preservative: 0.1% Sodium Azide. Dye: Violet (Acid Red No. 33 and Acid Blue No. 9).

- 1 bottle (1.5 mL) REAGENT B. Recalcified Plasma (Human) nonreactive for HBsAg, anti-HBs, anti-HCV, and anti-HIV-1/HIV-2. Preservative: 0.1% Sodium Azide. Dye: Yellow (Acid Yellow No. 23).

- 1 bottle (18 mL) DILUTION REAGENT. Recalcified Plasma (Human) nonreactive for HBsAg, anti-HBs, anti-HCV and anti-HIV-1/HIV-2.
  Preservative: 0.1% Sodium Azide.

III. MANUFACTURING AND CONTROLS

A. Manufacturing and Controls

Abbott MEIA Test System is prepared under U.S. License Number 43 by Abbott Laboratories. The hybridoma cell lines which produce antibodies reactive with HBsAg subtype Ad/Ay determinants were generated by fusing spleen cells from a BALB/C Mouse immunized with purified HBsAg subtype Ad and Ay with the mouse myeloma cell line SP2/0 using a modification of the procedures described by Kohler and Milstein (Eur. J. Immunol. (1976) 6, 511-9). The cell lines were expanded in medium and maintained in a tissue culture system. This material is purified and coupled to microparticles.

Positive and Negative Controls are prepared from human plasma, which are positive and negative, respectively, for HBsAg. The positive plasma is heated at 60°C for a minimum of 10 hours but complete inactivation may not have occurred. Reagent A is prepared from human plasma which is reactive for anti-HBs.

Raw materials intended for use in the product are subjected to appropriate quality control evaluations before they are accepted for use in manufacturing. Acceptance criteria and performance specifications have been established for all test kit components. Components are assembled into test kits, each lot of which is subjected to a final performance test.

Each lot of IMx HBsAg is tested with an in-house panel of samples with varying levels of HBsAg reactivity as well as the CBER HBsAg Reference Panel, and must meet the performance requirements of both panels.

The cell line used for this product is the same cell line used in Auszyme Monoclonal (PLA Ref. Nos. 84-385, 86-0021, and 86-0397) which is presently licensed by CBER.

B. Stability Studies

The stability of IMx HBsAg has been established based upon testing at the recommended storage conditions of 2 to 8°C and temperature extremes. Five lots of product were evaluated after being stored at 2 to 8°C for thirteen months. Four lots of product were each evaluated after being subjected to 37°C for 12 days and then returned to 2-8°C. The studies indicate no compromise in product performance to date and support a twelve month dating period for the test kit.

Two lots of Confirmatory Reagent A were each evaluated after being subjected to 2-8°C, 37°C and 45°C storage. These studies indicate no compromise in product performance and support a twelve month dating period for the Confirmatory kit.

C. Methods of Validation

Production of test kit components is monitored by in process testing. Product purity and potency are assured through evaluation of product appearance, sterility or bioburden tests and performance. Product performance is assessed through laboratory evaluations of each test kit against an in-house panel and the CBER HBsAg Reference Panel.

Each lot of product and protocol summarizing pertinent product testing are submitted for evaluation and approval by FDA prior to release for distribution.

D. Labeling

The product labeling, including immediate container and package labels and the package insert (directions for use), have been reviewed for compliance with 21 CFR 610.60, 610.61, 610.62 and 809.10 and found to be satisfactory. The package insert states that IMx HBsAg is a qualitative third generation Microparticle Enzyme

Immunoassay for the detection of Hepatitis B Surface Antigen (HBsAg) in human serum or plasma. The product trade name, IMx® HBsAg, is not known to conflict with any other biologic or device trade name.

The package insert states that the IMx HBsAg Confirmatory assay is for confirmation of the presence of Hepatitis B Surface Antigen (HBsAg) in human serum or plasma by means of specific antibody neutralization.

E. Establishment Inspection

A prelicensing inspection of the areas where product is manufactured, tested, stored and shipped was conducted on 6/14-6/18/93. Facilities and procedures were found to comply with current good manufacturing practices (CGMP). Subsequent inspections were also performed in June, 1994, and in January, 1995, and no violations of CGMP were found for the IMx® HBsAg or IMx® HBsAg Confirmatory assays.

F. Environmental Impact Analysis Report (EIAR)

A detailed EIAR was filed by the manufacturer. This product has no significant environmental impact. A summary of the procedures taken by the manufacturer to protect the environment are stated as follows.

1. Positive control human serum/plasma is heated at 60°C for a minimum of 10 hours before being used to manufacture the Positive Control.

   All biohazardous waste material disposed of as if it contains infectious agents.

2. Solution preparation, component manufacturing and labeling/packaging operations located in Buildings [redacted] will generate primarily aqueous process waste streams and washwaters from equipment preparation and cleanup. Wastewaters from these operations will be discharged directly to a publicly owned advanced tertiary - level activated sludge wastewater treatment plant for biological, chemical, and physical stabilization. All solid wastes will be managed in accordance with applicable State and Federal regulations.

G. Failure Mode Effects and Criticality Analysis (FMECA)

A failure mode effects and criticality analysis was performed on the screening and confirmatory tests addressing reagent volumes and incubation times and temperatures. System performance was assessed with each test situation. The conclusion drawn was that appropriate safeguards are designed into the system or are addressed in the package insert.

IV. BIOLOGICAL PRINCIPLES OF THE TEST

IMx® HBsAg is based on the Microparticle Enzyme Immunoassay (MEIA) technology. The IMx HBsAg reagents and sample are added to the reaction cell in the following sequence:

- The probe/electrode assembly delivers the sample and Anti-HBs (Mouse, Monoclonal) Coated Microparticles and Biotinylated Anti-HBs (Goat) solution to the incubation well of the reaction cell.
- The HBsAg binds to the Anti-HBs Coated Microparticles and Biotinylated Anti-HBs forming an antibody-antigen-antibody complex.
- An aliquot of the reaction mixture containing the antibody-antigen-antibody complex bound to the microparticles is transferred to the glass fiber matrix. The microparticles bind irreversibly to the glass fiber matrix.
- The Anti-Biotin: Alkaline Phosphatase Conjugate is dispensed onto the matrix and binds with the antibody-antigen-antibody complex.
- After washing to remove unbound materials, the substrate, 4-Methylumbelliferyl Phosphate (MUP), is added to the matrix. The alkaline phosphatase (AP)-labeled conjugate catalyzes the removal of a phosphate group (H_3PO_4) from the substrate, yielding the fluorescent product, 4-methylumbelliferone (MU). This fluorescent product is measured by the MEIA optical assembly.

$$MUP + H_2O \xrightarrow{AP} MU + H_3PO_4$$

The presence or absence of HBsAg is determined by comparing the rate of formation of fluorescent product to the Cutoff, which is calculated from the MODE 1 Calibrator rate. If the rate of the specimen is greater than or equal to the Cutoff, the specimen is considered reactive for HBsAg.

The IMx® HBsAg Confirmatory assay is based on the Microparticle Enzyme Immunoassay (MEIA) technology. The IMx HBsAg Confirmatory assay (Assay #74) uses the same reagents (IMx HBsAg Reagent Pack, No. 2228-25) as the IMx HBsAg assay. For information on the IMx HBsAg assay procedure, refer to the IMx HBsAg assay package insert.

The IMx HBsAg Confirmatory assay differs from the IMx HBsAg assay in that the specimen is preincubated with the IMx HBsAg Confirmatory Reagent A [a high titer anti-HBs (Human) Solution] or Reagent B [Recalcified Plasma (Human), nonreactive for

HBsAg, anti-HBs, anti-HCV and anti-HIV-1/HIV-2] prior to running the assay. If HBsAg is present in the specimen, it will be bound by Reagent A. The neutralized HBsAg is subsequently blocked from binding to the antibody coated microparticles.

The assay principle involves 2 steps: an off-line neutralization, and the automated running of the assay.

- The neutralizing step consists of manual, precision pipetting of each sample (including MODE 1 Calibrator, Positive Control, and specimens) into the sample well of two reaction cells. Reagent A is added to one reaction cell and Reagent B is added to the other. (See IMx HBsAg Confirmatory Procedure, Carousel Configuration).

- The probe/electrode assembly delivers the sample/Confirmatory Reagent mixture, Anti-HBs (Mouse, Monoclonal, IgM) Coated Microparticles, and Biotinylated Anti-HBs (Goat,-IgG) solution to the incubation well of the reaction cell.

- The available HBsAg binds to the Anti-HBs Coated Microparticles and Biotinylated Anti-HBs forming an antibody-antigen-antibody complex.

- An aliquot of the reaction mixture containing the antibody-antigen-antibody complex bound to the microparticles is transferred to the glass fiber matrix. The microparticles bind irreversibly to the glass fiber matrix.

- The Anti-Biotin (Rabbit): Alkaline Phosphatase Conjugate is dispensed onto the matrix and binds with the antibody-antigen-antibody complex.

- After washing to remove unbound materials, the substrate, 4-Methylumbelliferyl Phosphate (MUP), is added to the matrix. The alkaline phosphatase (AP)-labeled conjugate catalyzes the removal of a phosphate group (H_3PO_4) from the substrate, yielding the fluorescent product, 4-methylumbelliferone (MU). This fluorescent product is measured by the MEIA optical assembly.

$$MUP + H_2O \xrightarrow{AP} MU + H_3PO_4$$

V. CLINICAL DATA

A. Non-Clinical (In-house) Data

1. In addition to the clinical studies, in-house (non-clinical) studies were done on high risk populations. Twenty-five homosexual males and 10 specimens reactive for HCV were tested. Of the 35 specimens tested, only one specimen from a homosexual male was

found to be reactive. It was confirmed HBsAg positive by neutralization with human anti-HBs.

2. Three patient serial bleed panels were also studied. The onset of HBsAg, rise and decline in titer in each panel was detected with 100% agreement between IMx HBsAg and Auszyme® Monoclonal.

3. A study was performed to determine the minimum and maximum size of an assay run. The purpose of this study was to demonstrate that no front to back effects are present and that the same results are obtained for a minimum size run as for a maximum size run. The studies showed the acceptable batch size range may vary from 1-20 specimens for a Calibration Assay or 1-21 specimens for a Mode 1 Assay.

4. Studies were performed to validate the specimen shipping and handling conditions delineated in the package insert. Reactive and nonreactive specimens were drawn into seven different anticoagulant tubes as well as serum and serum separator tubes. These specimens were subjected to freeze/thaw cycles, 14 day storage at 2-8°C, and a ship stress. The studies demonstrated that all tested tube types were acceptable for use in this assay, and that specimens may be shipped under ambient or refrigerated conditions, or frozen on dry ice.

   Two studies were performed to determine if biotin or its analogs interfered with the assay causing false positive or negative results. The conclusion drawn from these studies is that neither biotin nor its analogs cause interference for IMx HBsAg.

B. Summary of Clinical Data

Clinical studies were performed in 1989 and in 1992. The data presented represent both studies, as well as studies presented in Amendment #1. These data have been combined for inclusion in the package inserts.

1. Reproducibility (1989 studies)

Assay reproducibility was determined by assaying a 9 member proficiency panel in replicates of two in 10 consecutive runs at a total of four sites. Data from this study were recalculated to demonstrate intra-assay and inter-assay variability which is more consistent with the presentation of the October 1992 reproducibility study results.

ABBOTT IMx HBsAg REPRODUCIBILITY

PANEL MEMBER	N	MEAN S/N	INTRA-ASSAY SD	INTRA-ASSAY %CV	INTER-ASSAY SD	INTER-ASSAY %CV
1	140	- 1.886	0.061	3.3	0.098	5.3
2	140	2.087	0.055	2.6	0.110	5.3
3	140	2.625	0.083	3.2	0.147	5.6
4	140	4.667	0.148	3.2	0.243	5.2
5	136	1.608	0.051	3.2	0.081	5.0
6	140	2.260	0.074	3.3	0.115	5.1
7	140	2.718	0.072	2.7	0.136	5.0
8	140	5.372	0.196	3.7	0.329	6.1
9	140	1.015	0.045	4.4	0.055	5.4
MODE 1	70*	1.002	0.000	0.0	0.048	4.8
NC	140	0.982	0.037	3.8	0.050	5.1
PC	140	12.667	0.577	4.6	0.787	6.2

NC = Negative Control
PC = Positive Control
* = One replicate per run

2. Reproducibility (October 1992 studies)

In this study, six specimens were assayed in replicates of three in eight consecutive runs using three lots of reagents at a total of five sites. The intra-assay, inter-assay and inter-lot Standard Deviation (SD) and Percent Coefficient of Variation (%CV) were calculated. Mean Sample to Negative (S/N) is defined as the Mean Sample Rate divided by the MODE 1 Calibrator Rate.

PANEL MEMBER	N	MEAN S/N	INTRA-ASSAY SD	%CV	INTER-ASSAY SD	%CV	INTER-LOT SD	%CV
A	671	5.983	0.176	2.9	0.379	6.3	1.099	18.4
B	670	2.267	0.072	3.2	0.129	5.7	0.288	12.7
C	672	3.715	0.101	2.7	0.208	5.6	0.598	16.1
D	671	1.953	0.062	3.2	0.109	5.6	0.214	10.9
E	671	3.051	0.084	2.7	0.172	5.6	0.444	14.6
F	670	1.139	0.044	3.8	0.066	5.8	0.067	5.9
MODE 1	224*	0.994	0.000	0.0	0.056	5.7	0.058	5.8
NC	448	0.966	0.042	4.4	0.058	6.0	0.060	6.2
PC	447	16.192	0.457	2.8	0.974	6.0	3.859	23.8

NC = Negative Control
PC = Positive Control
* = One replicate per run

3. Reactivity

To determine the reactivity in random donor populations, a total of 7,485 serum and plasma specimens from volunteer blood donors was evaluated at five different U.S. sites. Surplus random samples which met the volume requirement of the clinical protocol were used for testing. Two sites evaluated 2,001 random, unlinked plasma specimens and had a total of zero (0.00%) repeatedly reactive specimens by IMx HBsAg. Four sites evaluated a total of 5484 random, unlinked serum specimens by IMx HBsAg and had a total of 12 (0.22%) repeatedly reactive specimens. The presence of HBsAg in the repeatedly reactive specimens was confirmed by neutralization with anti-HBs. Eleven (0.20%) of the repeatedly reactive serum specimens confirmed positive for the presence of HBsAg. One site evaluated a total of 966 random, unlinked plasmapheresis donor specimens, of which zero (0.00%) were repeatedly reactive by IMx HBsAg.

	IMx HBsAg		
1989 CLINICAL STUDIES NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Volunteer Blood Donors			
Plasma (1 site) Total 1000	0 (0.00%)	0 (0.00%)	0 (0.00%)
Serum (3 sites) Total 4484	15 (0.33%)	11 (0.25%)	11 (0.25%)
GRAND TOTAL 5484 (3 sites)	15 (0.27%)	11 (0.20%)	11 (0.20%)
1992 CLINICAL STUDIES NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Volunteer Blood Donors			
Plasma (1 site) Total 1001	2 (0.20%)	0 (0.00%)	NT*
Serum (1 site) Total 1000	1 (0.10%)	1 (0.10%)	0 (0.00%)
GRAND TOTAL 2001	3 (0.15%)	1 (0.05%)	0 (0.00%)
Plasmapheresis Donors (1 site) Total 966	0 (0.00%)	0 (0.00%)	NT*
COMBINED STUDIES NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Volunteer Blood Donors			
Plasma (2 sites) Total 2001	2 (0.10%)	0 (0.00%)	NT*
Serum (4 sites) Total 5484	16 (0.29%)	12 (0.22%)	11 (0.20%)
GRAND TOTAL 7485 (5 sites)	18 (0.24%)	12 (0.16%)	11 (0.15%)
Plasmapheresis Donors (1 site) Total 966	0 (0.00%)	0 (0.00%)	NT*

*NT = Not tested

To determine the reactivity in patient populations, six different sites evaluated a total of 3,594 specimens submitted to hospital and public health laboratories for diagnostic testing. Of these specimens, 101 (2.81%) were repeatedly reactive by IMx HBsAg. The presence of HBsAg was confirmed in 88 (2.45%) specimens by neutralization with anti-HBs. Six different sites evaluated 986 specimens from obstetrical/gynecological (OB/GYN) patients. Eighty-one (8.22%) specimens were repeatedly reactive by IMx HBsAg and 77 (7.81%) confirmed positive for the presence of HBsAg. Of the 77 confirmed positive specimens, 56 specimens were from an inner-city population with an HBsAg prevalence rate determined to be 28.57%.

	IMx HBsAg		
1989 CLINICAL STUDIES NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Health/Public Health Patients (5 sites) Total 3094	110 (3.56%)	96 (3.10%)	84 (2.71%)
OB/GYN (2 sites) Total 500	22 (4.40%)	20 (4.00%)	20 (4.00%)
1992 CLINICAL STUDIES NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Hospital/Public Health Patients (2 sites) Total 500	10 (2.00%)	5 (1.00%)	4 (0.80%)*
OB/GYN** (5 sites) Total 486	64 (13.17%)	61 (12.55%)	57 (11.73%)

* The volume of one specimen was insufficient for Confirmatory testing by IMx HBsAg.

** All specimens were from pregnant females.

COMBINED STUDIES NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Hospital/Public Health Patients (6 sites) Total 3594	120 (3.34%)	101 (2.81%)	88 (2.45%)
OB/GYN (6 sites) Total 986	86 (8.72%)	81 (8.22%)	77 (7.81%)

To determine the reactivity in selected populations with Hepatitis B, six different sites evaluated a total of 224 specimens from 224 individuals diagnosed with acute or chronic Hepatitis B. Four different sites evaluated 184 specimens that were unclassified as to disease state but previously classified as HBsAg positive. In 93 patients with acute Hepatitis B, 93 (100.00%) specimens were repeatedly reactive and 92 (98.92%) confirmed positive for the presence of HBsAg by neutralization. In 131 patients with chronic Hepatitis B, 131 (100.00%) specimens were repeatedly reactive and confirmed positive for the presence of HBsAg by neutralization. In 184 specimens of unknown clinical status, but previously HBsAg positive, 181 (98.37%) were repeatedly reactive by IMx HBsAg and confirmed positive for the presence of HBsAg.

	IMx HBsAg		
1989 CLINICAL STUDIES NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Acute (2 sites) Total 43	43 (100.00%)	43 (100.00%)	42 (97.67%)*
Chronic (2 sites) Total 82	82 (100.00%)	82 (100.00%)	82 (100.00%)
Unknown Clinical Status (1 site) Total 37	36 (97.30%)**	36 (97.30%)	36 (97.30%)
GRAND TOTAL 162 (3 sites)	161 (99.38%)	161 (99.38%)	160 (98.77%)

* One specimen was nonreactive by IMx HBsAg when retested at Abbott Laboratories and was not confirmed.
** One specimen was nonreactive by IMx HBsAg.

1992 CLINICAL STUDIES NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Acute (3 sites) Total 50	50 (100.00%)	50 (100.00%)	50 (100.00%)
Chronic (4 sites) Total 49	49 (100.00%)	49 (100.00%)	49 (100.00%)
Unknown Clinical Status (4 sites) Total 147	145 (98.64%)*	145 (98.64%)	145 (98.64%)
GRAND TOTAL 246 (4 sites)	244 (99.19%)	244 (99.19%)	244 (99.19%)

* Two specimens were nonreactive by IMx HBsAg.

	IMx HBsAg		
COMBINED STUDIES NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Acute[a] (5 sites) Total 93	93 (100.00%)	93 (100.00%)	92[c] (98.92%)
Chronic[b] (6 sites) Total 131	131 (100.00%)	131 (100.00%)	131 (100.00%)
Unknown Clinical Status (4 sites) Total 184	181[d] (98.37%)	181 (98.37%)	181 (98.37%)
GRAND TOTAL 408 (6 sites)	405 (99.26%)	405 (99.26%)	404 (99.02%)

[a] Acute Hepatitis B included patients with elevated liver enzymes and reactive for HBsAg and anti-HBc IgM. Alcohol, toxic, other viral or drug related etiologies were excluded.

[b] Chronic Hepatitis B included patients with initial and follow up serum specimens (at least 6 months between blood draws) with ALT levels greater than 2x upper normal limit unless liver biopsy indicated chronic hepatitis and reactive for HBsAg and HBeAg and/or anti-HBe. Alcohol, toxic, other viral or drug related etiologies were excluded.

[c] One specimen retested at Abbott Laboratories was nonreactive by IMx HBsAg.

[d] Three specimens were nonreactive by IMx HBsAg and AUSZYME. Specimen misclassification was suspected.

To determine the reactivity in selected populations, specimens from individuals with hepatitis, other liver diseases, potentially interfering substances and individuals at high risk were tested by IMx HBsAg. Six different sites evaluated a total of 344 specimens from individuals with hepatitis and other liver diseases. Sixteen (4.65%) specimens were repeatedly reactive by IMx HBsAg. The presence of HBsAg was confirmed by neutralization in 12 (3.49%) specimens.

Six different sites evaluated a total of 512 specimens from individuals at high risk of infection with Hepatitis B virus. This category included 186 intravenous drug users, 219 dialysis patients, 97 hemophiliacs and 10 homosexual males. There were 10 (1.95%) specimens that were repeatedly reactive by IMx HBsAg. The presence of HBsAg was confirmed in 9 (1.76%) specimens.

Seven different sites evaluated 376 specimens with potentially interfering substances and from individuals with other diseases. Twelve (3.19%) specimens were repeatedly reactive by IMx HBsAg and 6-(1.60%) confirmed positive for the presence of HBsAg.

	IMx HBsAg		
1989 CLINICAL STUDIES NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Hepatitis/Other Liver Diseases (2 sites) Total 148	1 (0.68%)	1 (0.68%)	0 (0.00%)
High Risk for HBV Infection (3 sites) Total 216	8 (3.70%)	6 (2.78%)	6 (2.78%)
Potentially Interfering Substances (2 sites) Total 96	3 (3.13%)	3 (3.13%)	3 (3.13%)
Amendment #1 NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Hepatitis/Other Liver Diseases (1 site) Total 49	9 (18.37%)	9 (18.37%)	8 (16.33%)
High Risk for HBV Infection (1 site) Total 136	2 (1.47%)	2 (1.47%)	1 (0.74%)
Potentially Interfering Substances (1 site) Total 47	2 (4.26%)	2 (4.26%)	2 (4.26%)

	IMx HBsAg		
1992 CLINICAL STUDIES NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Hepatitis/Other Liver Diseases (4 sites) Total 147	7 (4.76%)	6 (4.08%)	4 (2.72%)
High Risk for HBV Infection (3 sites) Total 160	2 (1.25%)	2 (1.25%)	2 (1.25%)
Potentially Interfering Substances (5 sites) Total 233	10 (4.29%)	7 (3.00%)	1 (0.43%)
COMBINED STUDIES NUMBER TESTED	INITIALLY REACTIVE	REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Hepatitis/Other[a] Liver Diseases (6 sites) Total 344	17 (4.94%)	16 (4.65%)	12 (3.49%)
High Risk for HBV[b] Infection (6 sites) Total 512	12 (2.34%)	10 (1.95%)	9 (1.76%)
Potentially Interfering Substances[c] (7 sites) Total 376	15 (3.99%)	12 (3.19%)	6 (1.60%)

[a] Hepatitis/Other Liver Diseases includes Hepatitis A, NANB Hepatitis, Recovering Hepatitis B, Primary Biliary Cirrhosis, Primary Sclerosing Cholangitis, Hepatocellular Carcinoma, Drug-induced Liver Disease, Alcoholic Liver Disease, Hemochromatosis, Alpha-1 Antitrypsin Deficient, Autoimmune Hepatitis and other diseases (not specified).

[b] Individuals at High Risk includes Intravenous Drug Users, Dialysis Patients, Hemophiliacs and Homosexual Males.

[c] Potentially Interfering Substances includes specimens from patients diagnosed with Systemic Lupus Erythematosus, Rheumatoid Arthritis, Gammopathies, Syphilis, Toxoplasmosis, Positive for antibody to HSV, Rubella, CMV, EBV, HIV-1, HTLV-I, or Smooth Muscle.

C. Performance Characteristics

Specificity

The specificity of IMx® HBsAg was 99.99% (7,473/7,474*) in volunteer blood donors and 100.00% (966/966) in plasmapheresis donors based on an assumed zero prevalence of HBsAg.

* In these calculations 11 of the 12 repeatedly reactive volunteer blood donor specimens were excluded due to confirmation by neutralization with anti-HBs.

Sensitivity

The sensitivity of IMx HBsAg was evaluated by two different criteria.

1. The ability to detect HBsAg by IMx HBsAg compared to that of AUSZYME in patients with acute and chronic Hepatitis B and in specimens of unknown clinical status but previously classified as HBsAg positive.

1989 CLINICAL STUDIES NUMBER TESTED	IMx HBsAg REPEATEDLY REACTIVE	AUSZYME REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Acute Total 43	43 (100.00%)	43 (100.00%)	42 (97.67%)
Chronic Total 82	82 (100.00%)	82 (100.00%)	82 (100.00%)
Unknown Clinical Status, Total 37	36 (97.30%)	36 (97.30%)	36 (97.30%)
GRAND TOTAL 162	161 (99.38%)	161 (99.38%)	160 (98.77%)

1992 CLINICAL STUDIES NUMBER TESTED	IMx HBsAg REPEATEDLY REACTIVE	AUSZYME REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Acute Total 50	50 (100.00%)	50 (100.00%)	50 (100.00%)
Chronic Total 49	49 (100.00%)	49 (100.00%)	49 (100.00%)
Unknown Clinical Status, Total 147	145 (98.64%)	144 (97.96%)	145 (98.64%)
GRAND TOTAL 246	244 (99.19%)	243 (98.78%)	244 (99.19%)

COMBINED STUDIES NUMBER TESTED	IMx HBsAg REPEATEDLY REACTIVE	AUSZYME REPEATEDLY REACTIVE	CONFIRMED POSITIVE
Acute Total 93	93 (100.00%)	93 (100.00%)	92 (98.92%)
Chronic Total 131	131 (100.00%)	131 (100.00%)	131 (100.00%)
Unknown Clinical Status Total 184	181 (98.37%)	180 (97.83%)	181 (98.37%)
GRAND TOTAL 408	405 (99.26%)	404 (99.02%)	404 (99.02%)

2. The sensitivity of IMx HBsAg was evaluated in the 1992 Clinical Studies by assaying a sixteen member panel of purified HBsAg (Ad and Ay) specimens. Each panel member was assayed in replicates of two using three lots of reagents at a total of two sites.

Detection of Purified HBsAg/Ad by IMx HBsAg

Concentration (ng/mL)	S/N Value	Result
2.18	5.94	+
1.51	4.42	+
0.92	3.04	+
0.77	2.73	+
0.49	2.26	+
0.37	1.98	-
0.23	1.70	-
0.10	1.44	-

Detection of Purified HBsAg/Ay by IMx HBsAg

Concentration (ng/mL)	S/N Value	Result
2.30	7.29	+
1.59	5.50	+
0.92	3.50	+
0.84	3.26	+
0.62	2.59	+
0.45	2.20	+
0.29	1.86	-
0.17	1.51	-

D. CONFIRMATORY RESULTS

Specific Performance Characteristics

1989 CLINICAL STUDIES

HBsAg Confirmatory for Auszyme Monoclonal	IMx HBsAg Confirmatory +	IMx HBsAg Confirmatory −
+	60	0
−	0	0

n=60

1992 CLINICAL STUDIES

HBsAg Confirmatory for Auszyme Monoclonal	IMx HBsAg Confirmatory +	IMx HBsAg Confirmatory −
+	242	0
−	1	0

n=243

COMBINED STUDIES

HBsAg Confirmatory for Auszyme Monoclonal	IMx HBsAg Confirmatory +	IMx HBsAg Confirmatory −
+	302	0
−	1*	0

n=303

* Specimen retested at Abbott Laboratories was confirmed positive for HBsAg by HBsAg Confirmatory for Auszyme Monoclonal.

VI. PACKAGE INSERTS

A copy of each of the following package inserts is attached:

- IMx® HBsAg
- IMx® HBsAg Confirmatory

Licensing Review Commitee:

Leonard Wilson, Chairperson

Janet Claggett

Robin Biswas, M.D.